EFFECTS OF pH, ENZYMES, AND STORAGE TIME ON THE RHEOLOGY OF SWEET POTATO PUREE

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-ABSTRACT-

Sweet potato purees were pH adjusted (some also heat treated) and stored up to 9 months in sealed containers at room temperature. Some purees were treated with amylolytic enzymes before or after storage. Rheology and carbohydrates were evaluated after 0, 3, 7, and 9 months. Nonenzyme treated purees at 4.2 pH were semi-solids after 3 months storage, but those at 1.5 and 11.5 pH had lower viscosities. Amylolytic enzyme treatments lowered molecular weights and viscosities. Pre-storage treatments were more effective than post storage in molecular weight reduction. Post storage treatments were more effective in viscosity reduction.

INTRODUCTION

SWEET POTATOES are potentially a good source of carbohydrates, proteins, and vitamins suitable for food and industrial uses (Purcell et al., 1972). However, length of growing season and storage problems, involving energy use and decay, prevent realization of the potential in this crop. Techniques presently used for storage are inadequate to extend the operating season of processing plants (Ice, 1978); therefore, this, and related studies were initiated to explore methods of increasing the usability of sweet potatoes through processing.

One such method is to store sweet potatoes as a microbiologically stabilized puree. Pureeing has been a method of preparing several crops for storage, and the purees—either cooked or raw—are stored in large containers for further processing into various products (Anon., 1976; Wilson, 1976). In previous applications of this type of technology, materials of low starch and high water content were used; thus, concern about flow characteristics of the puree was reduced. Use of materials high in acid content also reduces the need for extensive thermal processing (NCA, 1973).

Plant tissue purees generally have pseudoplastic flow characteristics and a yield stress (Muller, 1973). Under certain conditions, such as heat or NaOH treatment, starch or pectin gels can form and increase both the yield stress and the apparent viscosity of the puree such that it will act more like a solid. If a commodity is to be pureed, flow characteristics of the puree should be investigated so that it might be prepared and handled expeditiously. These characteristics would not only determine the kinds and amounts of chemicals to be included in the puree, but also the penetration of heat into the puree.

The technology used before storage should be simple and rapid to minimize handling. If the sweet potato puree were fluid and had a relatively low yield stress, it could be

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pumped into and out of storage tanks.

The viscosity of sweet potato puree would be expected to depend on the carbohydrate fraction (Heckman, 1977). If the starch gelatinizes the puree will increase in viscosity and may even solidify. Heat treatment or solubilization of starch with NaOH would change the form of the starch from granular to diffuse and the change would markedly increase the viscosity of the puree (Heckman. 1977). Hydrolysis of starch would be expected to reduce viscosity by reducing the sizes of the starch molecules (Whitaker, 1972); but, pectin could play a role (Nelson et al., 1977) and thus, complicate the problem.

The present study was to develop data on the flow characteristics of sweet potato purees prepared and preserved by different methods.

EXPERIMENTAL

THE VARIABLES in this study were microbiological stabilization treatments, amylolytic enzyme treatments to hydrolyze starch, and storage times (Ice, 1978). Microbiological stabilization involved pH adjustment with or without pasteurization. The enzyme treatments included no enzyme addition, prestorage enzyme treatment and post storage treatment. Storage times were for 0, 3, 6, 7, and 9 months at room temperature (18-25°C).

Freshly harvested Jewel cultivar sweet potatoes were comminuted with a Fitz Mill (Fitzpatrick Co., Chicago, Ill.) to particles of 0.03 inches or smaller in diameter. Five percent by weight water and 3% by weight 0.25M CaCl₂ solution in water were added. Water was added to improve flowability of the raw puree, and CaCl, was added to increase Ca++ concentration of the puree to stabilize an α-amylase enzyme used later in processing some of the puree. The puree was divided into two lots. One lot was subdivided into three parts, and these were brought to pH 1.5, 4.2, and 11.5 with either concentrated H₃PO₄ or 10N NaOH. The portion acidified to pH 4.2 was also pasteurized at 90°C for at least 5 min. The three portions were canned and stored for different periods. Samples of each of these canned portions were evaluated and analyzed at specified times as described below. For the post storage treatment other samples from each of the three portions were brought to pH 5 with NaOH or H₃PO₄, treated with amylolytic enzymes, and analyzed. The second lot was enzymatically hydrolyzed and then divided into three parts. These were microbiologically stabilized like the first lot and then canned. Organization of the procedure is given in Tables 1

Table 1—Codes for identifying samples

Three-part codes that begin with a number

- A. The number at the start of the code indicates the pH of the samples (1.5, 4.2, and 11.5). Samples that were adjusted to pH 4.2 were also pasteurized for storage.
- B. The designations E0, E1, and E2 indicate no enzyme treatment, treatment before storage, and treatment after storage, respectively.
- C. The last part of the code indicates storage time in months. When all storage periods are meant, "X" is used.

Two-part codes

PRO:E0 indicates the raw puree with only the addition of water. PRO:E0 + Ca indicates PRO:E0 with the $CaCl_2$ solution added. GEL:E0 indicates PRO:E0 + Ca with heat gelatinization.

GEL:E1 indicates GEL:E0 which has undergone the enzyme treatment.

and 2. Dilution effects are listed in Table 2.

The canning procedure included filling clean glass (1 qt) containers with puree, filling the headspace with toluene, and sealing containers with enamel-lined lids. Only the samples at pH 4.2, which were pasteurized before canning, were packed hot.

For enzyme treatment, the puree was heated to 90°C by steam injection heating to gelatinize the starch. Next, an α -amylase preparation [Tenase® (Miles Laboratories, Inc.) used at a level of 1 ml per kg puree] was added to the puree to reduce its viscosity by randomly hydrolyzing bonds within the starch molecules. The puree was then allowed to cool slowly at room temperature. This attack on starch increased the number of carbohydrate molecules and, hence, the number of carbohydrate end-groups, which were hydrolyzable by β -amylase and amyloglucosidase. Without inactivating either the natural or supplementary α -amylase, we added β -amylase [Clarase® (Miles Laboratories, Inc.)] and amyloglucosidase [Diazyme® (Miles Laboratories, Inc.)] at levels of 0.06g and 0.01 ml per kg of puree, respectively, and then incubated the puree for about 48 hr at 55°C (Ice, 1978).

At completion of the specified storage periods, samples (E0 and E1) were examined as to general physical character. Containers were checked for etching, and closures were checked for corrosion and other changes. Vacuum or pressure inside the containers was measured by a penetrating gauge, and the pH of the contents was determined. Color, texture, and odor of the samples were evaluated subjectively. Viscometric tests and carbohydrate analyses were made for all samples, including those in the E2 series. Some of the 4.2:E0:X samples were treated with a pectinase enzyme to observe if gelation was at least in part due to pectin.

A collateral study was made with sweet potatoes which had cured naturally incubating at about 29°C and 90% RH for 15 days. These cured sweet potatoes were processed and tested like the freshly harvested ones, except that only one storage time (7 months) was tested. The results were compared with the corresponding results for the uncured sweet potatoes to determine whether curing affected the quality of the stored puree.

Instrumentation

Three different instruments were used to measure, in duplicate,

Table 2—Samples and dilutions caused by additions of reagents during treatments

Sample	Starting material ^a	Additions	% Sweet potato material, by wt
PRO:E0	Sweet Potato	Water	95
PRO:E0 + Ca	PRO:E0	CaCl ₂ solution	92
1.5:E0:X	PRO:E0 + Ca	H ₃ PO ₄ to lower pH to 1.5	68
1.5:E2:X	1.5:E0:X	NaOH to raise pH to 5 after storage, enzyme preparations	50
4.2:E0:X	PRO:E0 + Ca	H ₃ PO ₄ to lower pH to 4.2, pasteurize	91
4.2:E2:X	4.2:E0:X	NaOH to raise pH to 5 after storage, enzyme preparations	91
11.5:E0:X	PRO:E0 + Ca	NaOH to raise pH to 11.5	86
11.5:E2:X	11.5:E0:X	H ₃ PO ₄ to lower pH to 5 after storage,	
		enzyme preparations	84
GEL:E0	PRO:E0 + Ca	Steam injection	92
GEL:E1 1.5:E1:X	GEL:E0 GEL:E1	Enzyme preparations H ₃ PO ₄ to lower pH	92
		to 1.5	70
4.2:E1:X	GEL:E1	H ₃ PO ₄ to lower pH to 4.2, pasteurize	
11.5:E1:X	GEL:E1	NaOH to raise pH to 11.5	88

^a Material in this column plus material in additions column are constituents of sample column, i.e., 1.5:E0:X is composed of PRO:E0 + Ca plus H₃PO₄.

the rheological properties of the purees. A Brookfield Synchro-Lectric Viscometer, Model RVT (Brookfield Engineering Laboratory, Inc.) was used during processing to determine the effect of α -amylase hydrolysis on the viscosity of the starch gel. This instrument was also used to obtain static yield stress values and stress values at 5 rpm for purees after storage treatment. We used a Rotovisco, Model RVI (Haake, Inc.) to obtain viscosity data at different shear rates and then developed flow characteristic curves from those data. An Instron Universal Testing Instrument, Table Model 1130 (Instron Corp.) was used with extrusion devices when the samples were too viscous to be tested with the Haake instrument.

The Brookfield instrument was used with a spindle consisting of a fluted metal cylinder 2 cm in height and 0.8 cm diameter. The surface of the cylinder had 12 flutes 0.05 cm deep and 0.05 cm wide oriented parallel to the axis of the rotation to prevent formation of a water layer which would allow the puree to slip. The rotational speed of the Brookfield instrument could not be converted to shear strain rate, because this test was run under wide rotational gap conditions. The width of the gap was variable because it depended on the yield stress. Yield shear stress was obtained from the same test that gave the steady state shear stress at 5 rpm. It was calculated from the maximum reading on the torque scale before rotation started.

The Haake Rotovisco instrument was used with three different rotors—Rotor SVIIP (height 1.96 cm, diameter 2.02 cm), rotor MVI (height 6.00 cm, diameter 4.01 cm), and rotor MVII (height 6.00 cm, diameter 3.68 cm). Different cups were used with the rotors, and the gaps between rotor and cup surfaces differed. Narrow gap conditions were maintained, so the gap was constant for a given rotor-cup combination; thus, shear strain rates could be calculated. The SVIIP rotor was fluted when manufactured, and the other rotors were fluted later to prevent separation of a water layer and, therefore, misleading readings. The data were converted to shear stress, shear rate, and dynamic yield stress (Rao, 1974). These data formed the basis of flow characteristic curves, which were plots of shear stress versus strain rate.

The Instron Machine was used with two extrusion rheometers, each of which was equipped with a piston that fitted closely into a cup. Forcing the piston into the cup when it was filled with puree caused the puree to flow through a small discharge cylinder attached to the cup. The diameter of the cup in one rheometer was 5.72 cm, and the attached discharge cylinder was 10.00 cm long and 0.476 cm in diameter. The cup diameter of the other rheometer was 3.80 cm; and the discharge cylinder was 10.60 cm long and 0.3173 cm in diameter. Force required to extrude the puree was recorded. Piston velocities varied from 3.33×10^{-4} to 3.33×10^{-3} m/s. These data were converted to shear stress and shear strain rate, and were plotted the same way that data from the rotational viscometers were plotted.

In all viscometry, except where specifically stated, the samples were tested at room temperature, i.e., $16-25^{\circ}$ C. The rheological properties of the samples were not temperature dependent within this range.

Treatment of viscosity data

In general, plots of shear stress versus shear strain rate fit a curve represented by the following equation:

$$\tau = \tau_0 + k \gamma^n \tag{1}$$

where τ is shear stress; τ_0 is yield stress; $\hat{\gamma}$ is shear strain rate; and k and n are constants. We calculated the constants n, k and τ_0 from rotational viscometer data, using appropriate equations from standard references (i.e. Van Wazer et al., 1963). Details are given in Ice (1978).

Because of our inability to measure the extent of plug flow (solid behavior at the center), data from extrusion rheometers were fit to the following simple power (equation) which does not include the yield stress:

$$\tau = k \dot{\gamma}^n \tag{2}$$

For statistical analysis of combined data from both the Haake and extrusion instruments, the latter, simpler equation was used.

Because the values of n and k varied inversely, and because very similar curves could be produced by different pairings of n and k, the product of these terms was calculated when Eq (2) was the model used. If Eq (2) is differentiated, the result is

$$\frac{\mathrm{d}r}{\mathrm{d}\dot{\gamma}} = nk\dot{\gamma}(n-1) \tag{3}$$

If $\dot{\gamma}$ is set equal to 1, the term $\dot{\gamma}^{(n-1)}$ is equal to 1; and therefore, can be dropped from Eq (3). The result is

$$\frac{d\tau}{d\hat{\gamma}} = nk$$

$$\hat{\gamma} = 1$$
(4)

The product nk is then the slope of τ versus $\dot{\gamma}$ at a strain rate of unity. We used nk to compare fluids since it was a good approximation of the initial slope of the τ versus $\dot{\gamma}$ curve for the data collected, assuming no yield stress (Fig. 1).

An apparent viscosity of the puree was determined at a shear strain rate of 200 s⁻¹ by interpolation. This shear strain rate was chosen because it represented a point on the nearly linear part of the shear stress versus shear strain rate curve (Fig. 1).

Yield stress (Fig. 1) was obtained by two methods. Static yield stress was derived from the torque required to induce shearing in the puree with the Brookfield instrument. We obtained dynamic yield stress from a curve that could be expressed by Eq (1) (Rao, 1974). In most instances, the curve of best fit intercepted the shear stress axis, and the intercept equalled the yield stress.

Carbohydrate data

Of the various carbohydrates in raw sweet potato, starch is the predominant one. Therefore, the state of the starch would be expected to strongly affect the flow character of the puree. We measured the amounts of total carbohydrate that could be extracted from the purees with 80% ethanol (Dubois et al., 1956) and used the values as indicators of the amount of starch that had degraded. All carbohydrate measurements were made in duplicate.

Statistical analysis

A three-way analysis of variance was used to indicate the relationships of the independent variables (enzyme treatment, storage time, storage pH) and dependent variables (apparent viscosity at a shear rate of 200 s⁻¹, yield stress, and nk). The nk values were from curves expressing Eq (2). The values for carbohydrate soluble in 80% ethanol were compared with the rheological data.

The design of this experiment (Tables 1 and 2) called for comparisons between the post storage enzyme treatment and the prestorage enzyme treatment and between the prestorage enzyme treatment and no enzyme treatment.

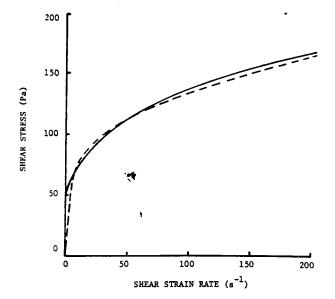


Fig. 1—Representation of the flow character of a specific puree by Eq. (2) (———): n = 0.253, k = 43.6; and Eq. (1) (———): n = 0.454, k = 11.0, and $\tau_0 = 50.2$.

The analysis of data for comparisons at 7 months' storage of purees made from cured and uncured roots was made with Student's t-test (Snedecor and Cochran, 1967).

RESULTS & DISCUSSION

General evaluation of containers, closures, and purees

The containers showed no damage from any of the treatments. The caps were sound in all instances; however, in samples at pH 11.5 stored 9 months without enzyme treatment the enamel lining had begun to turn green. Because toluene filled the headspace, this was not due to direct contact between cap and puree.

Measurement of internal pressure made before opening containers showed a vacuum in all instances, indicating a lack of growth of gas-producing microorganisms. The vacuum readings could be separated into two general groupings. First were those samples which were pasteurized and hot filled into containers, both with and without enzyme treatment (the pH 4.2 samples). Vacuums in these series of samples ranged from 4.7×10^4 to 6.1×10^4 Pa (14-18) inches of mercury). The second grouping of vacuums included all other samples. The vacuums of these samples varied somewhat -3.4×10^3 to 3.0×10^4 Pa (1-9) inches of mercury)—because the containers were filled and sealed before the puree had equilibrated to ambient temperature.

Samples at pH 1.5 stored 6 months without enzyme treatment gelled and could be rotated as a unit within the container. The static yield stress averaged 325 Pa and was higher than the average for this series (Table 3). The puree did not flow easily with gravity. The sample series at pH 1.5 pretreated with enzymes had a smoother appearance than the series without enzyme treatment.

Graininess was apparent in the sample series at pH 11.5 without enzyme treatment and the static yield stress was higher than could be measured with the Brookfield RVT instrument (Table 3). Some of the cellular structures of the tissue were retained intact. The series at pH 11.5 with an enzyme pretreatment was smoother and much less viscous than the series without enzyme treatment.

All samples at pH 4.2 had a smoother appearance because of the heat treatment involved in pasteurization or gelatinization. Samples adjusted to pH 4.2 before storage were highly viscous when being filled into containers at 80-90°C; and over a period of 3 months those without added enzymes-4.2:E0:X-became the strongest gels in the experiment. Their static yield stresses were much greater than could be measured with the Brookfield RVT instrument. The sample series at pH 4.2 with enzyme pretreatment poured easily during filling, but these samples were also gelled by the end of 3 months' storage. These gels were much weaker than the gels in series at pH 4.2 without

Table 3—Average static yield stress values and standard deviations for the three series

Average static yield stress (Pa)			
1.0	249	(80)	
	258	(140)	
	31	(12)	
Much greater than could be evaluateda			
	225	(21)	
	169	(88)	
Greate	r than could be	evaluated (approx	x. 400)
	38	(17)	
	110	(54)	
	. н М и	249 258 31 Much greater than 225 169 Greater than could be 38	249 (80) 258 (140) 31 (12) Much greater than could be evaluate 225 (21) 169 (88) Greater than could be evaluated (approx 38 (17))

^a The maximum static yield stress that could be measured accurately with the apparatus used was 358 Pa.

enzyme treatment. The series at pH 4.2 without enzyme treatment and with pretreatment discolored at the interface between the puree and toluene filling the headspace of the containers. This was caused by pigments leaching into the toluene. This phenomenon was not noted in the other series of samples.

Generally, addition of a large amount of acid darkened the puree noticeably. The NaOH addition also caused darkening. These color changes were reversible; for when the purees were brought to pH 5 for post storage treatment, they assumed a more nearly normal color.

At the end of storage, the pH of the most highly acidified series of samples varied somewhat (1.2-1.8), possibly because of the extreme acidity and its effect on the pH meter. Variations may also have been caused by changes in temperature during the processing of the puree. There was no noticeable variation in pH in the other series of samples.

Lack of appreciable variation between samples in a series was taken as an indication that acid producing microorganisms were not active in the puree. The presence of vacuum and lack of visual evidence of mold growth or other microbiological activity also indicated that the purees were stable through the storage periods. No off odors, which might have indicated spoilage, were noted. Additional support for the indication that these purees were microbiologically stable was the fact that the amount of carbohydrate did not change significantly with storage time (Ice, 1978).

Shear stress changes caused by α -amylase hydrolysis

Typical effects of the α -amylase hydrolysis over a period of 15 min are shown in Figure 2. Both static yield stress and stress at 5 rpm indicated that the enzyme preparation containing α -amylase effectively hydrolyzed gelatinized starch. The plots show that these parameters dropped dramatically over the first 5 min and then appeared to stabilize.

Material flow representations

When data from the Haake instrument were represented by Eq (1), which included the dynamic yield stress, correlation coefficients derived from curve fitting were generally in excess of 0.99. Specific yield stresses were reported by Ice (1978). When Eq (2) was used (no yield stress value), the correlation coefficients were somewhat reduced, but were generally in excess of 0.97. In only three instances were the r values between 0.90 and 0.94, and no correlation coefficient was lower than 0.90.

The apparent viscosity at $\dot{\gamma} = 200 \text{ s}^{-1}$ and nk [from Eq (2)] varied directly and almost identically (Fig. 3).

Effects of independent variables on flow characteristics

The enzyme treatments significantly reduced nk and apparent viscosity (Table 4), especially in the pH 4.2 samples. At that pH, the viscosity measurements of the E1 and E2 samples were less than one-third those of the E0 samples (Fig. 3). When gels from the sample series at pH 4.2 without enzyme treatment were subsequently exposed to enzymes which act on pectins, the purees became less solid. When these samples were treated with α -amylase to form the post storage enzyme-treated samples the gels were also broken. These observations indicate that both starch and pectin contributed to gel formation. The higher average viscosity for samples at pH 4.2 with enzyme pretreatment over the series with enzyme post treatment (Fig. 3) may have been due to pectin gel formation. The post storage enzyme-treated series was measured soon after enzyme hydrolysis and may not have had time to form a pectin gel.

The nk average and apparent viscosity data show that only in the 11.5 pH series was the pre-storage enzyme treatment more effective in reducing viscosity than the post storage enzyme treatment (Fig. 3). This lower viscosity

with pre-storage enzyme treatment may be indicative of the effects of the enzymes in the absence of pectic effects. The pH was too high for pectin gel formation (Nelson et al., 1977), and the purees were only moderately diluted with reagents.

Statistically, storage pH had a highly significant effect on nk and apparent viscosity. The high level of significance was largely due to the high nk and apparent viscosity values for the sample series stored at pH 4.2 without enzyme

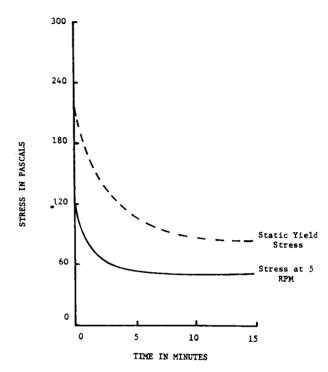


Fig. 2—Effect of α -amylase on the strength of starch gel at 90° C as measured with the Brookfield viscometer.

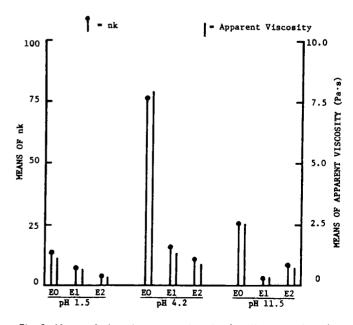


Fig. 3—Means of nk and apparent viscosity for all storage times for each enzyme and pH treatment.

E0 vs E1					Level of Significance
Ethanol Soluble Carbohydr (lower in E0 than E1)	ate				0.01
Apparent Viscosity					0.05
(lower in E1 than E0)			1	1.	0.05
(lower in E1 than E0)	.:	*.*	1.12422		0.01

E1 vs E2	Level of Significance		
Ethanol Soluble Carbohydrate (lower in E2 than E1)		0.05	
Apparent Viscosity (lower in E1 than E2)		NSª	
nk (lower in E1 than E2)		NSª	

a Note from Fig. 3 that at pH 1.5 and 4.2 E2<E1 but this is reversed at pH 11.5.

treatment which had formed gels. Other contributing causes were the dilution effect of the reagents and the effects of the reagents on the starch. Storage time had no statistically significant effect on any of the dependent variables.

Effects of independent variables on carbohydrates

To better understand the cause of rheological changes, we evaluated our data on carbohydrates extracted in 80% ethanol. The concentration of total soluble carbohydrates (expressed in terms of undiluted puree) tended to increase with storage time and was greater for the hydrolyzed than for the unhydrolyzed samples (Fig. 4). However, the increase in concentration with time was not statistically significant. Pretreatment was more effective than post storage treatment with enzymes in increasing the concentration of the ethanol-soluble carbohydrates (Table 4). Probably the ion concentration of the post storage enzyme-treated samples was high enough to slightly depress the activity of the amylolytic enzymes. Data for the enzyme-treated samples show that, with one exception, apparent viscosity and soluble carbohydrate concentration were inversely related (cf. Fig. 3 and 4). The direct relation between these two parameters in the samples at pH 1.5 was probably due to the greater dilution in the post storage enzyme treatment than prestorage treatment.

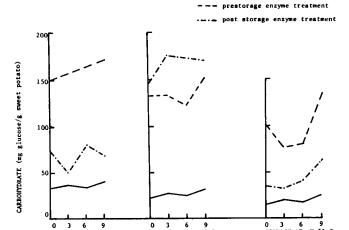
The values for total carbohydrate extracted with 80% ethanol are consistent with the fact that viscosity could be reduced by enzymatic hydrolysis. The enzymes may either reduce the amount of starch by converting it into sugars, or they may lower the average molecular weight of starch by breaking off polymeric units from the molecules as indicated by Muller (1971) and Heckman (1977).

Comparison between purees made with cured and uncured roots

Comparison on data on purees stored for 7 months showed that purees made from cured roots were slightly, but not significantly, lower in viscosity and starch content than those made from uncured sweet potatoes.

CONCLUSIONS

SWEET POTATO PUREE can be stored for 9 months at pH 1.5, 4.2 (+ pasteurization), or 11.5. The storage pH and/or enzyme treatment could determine the end use of the



no enzyme treatment

Fig. 4-Changes in carbohydrate soluble in 80% ethanol with months of storage.

puree, for it affects the extent of starch hydrolysis.

We believe that purees like those we prepared could be pumped into storage tanks, but that purees stored at pH 4.2 without enzyme treatment might not be pumped out of the storage tanks unless stirred with a device to overcome yield stress.

The enzymatic hydrolysis was effective in reducing viscosity. If pectin gel formation is likely, the enzyme system should include enzymes that act on pectin.

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